

Serial No. 09/914,279

IN THE CLAIMS:

1. (Currently amended) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

- (a) a metabolizable oil;
- (b) an emulsifying agent comprising an ionic detergent and a non-ionic detergent; and
- (c) at least one biologically active macromolecule selected from the group consisting of a polypeptide, a polynucleotide, a polynucleoside, an antigen, a pharmaceutical, a hormone, an enzyme, a transcription or translation mediator, an intermediate in a metabolic pathway, an immunomodulator, and an adjuvant,

wherein said biologically active macromolecule is adsorbed on the surface of the emulsion.

2. (Currently amended) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

- (a) a metabolizable oil; and
- (b) an emulsifying agent;

wherein said emulsifying agent comprises an ionic detergent and a non-ionic detergent, wherein said microemulsion is an oil-in-water emulsion comprising oil microdroplets, wherein substantially all of the oil microdroplets are less than 1 micron in diameter, and wherein said microemulsion exists in the absence of a polyoxypropylene-polyoxyethylene block copolymer.

3. (Previously presented) The microemulsion of claim 2, wherein said metabolizable oil is a member of the group consisting of an animal oil, an unsaturated hydrocarbon, a terpenoid, and a vegetable oil.

4. (Previously presented) The microemulsion of claim 3 wherein said metabolizable oil is a terpenoid selected from squalene, squalane and mixtures thereof.

5. (Previously presented) The microemulsion of claim 2 wherein said microemulsion comprises 0.5 to 20% by volume of said metabolizable oil in an aqueous medium.

Serial No. 09/914,279

6. (Currently amended) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

- (a) a metabolizable oil; and
- (b) an emulsifying agent;

wherein said emulsifying agent comprises an ionic detergent and a non-ionic detergent, and wherein said microemulsion comprises 0.01 to 2.5 % by weight of said emulsifying agent.

7. (Canceled)

8. (Previously presented) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

- (a) a metabolizable oil; and
- (b) an emulsifying agent;

wherein said emulsifying agent comprises an ionic detergent and wherein said emulsifying agent further comprises a non-ionic detergent selected from polyoxyethylene sorbitan monoesters, polyoxyethylene sorbitan diesters, polyoxyethylene sorbitan triesters, sorbitan monoesters, sorbitan diesters and sorbitan triesters.

9. (Currently amended) The microemulsion of claim 1 wherein said emulsifying agent ~~comprises~~ ionic detergent is a cationic detergent.

10. (Previously presented) The microemulsion of claim 9 wherein said cationic detergent is selected from the group consisting of hexadecyltrimethylammonium bromide, benzalkonium chloride, dimethyl dioctadecyl ammonium bromide, dioleoyl-3-trimethylammonium-propane, dodecyltrimethylammonium bromide, benzyldimethylhexadecyl ammonium chloride, cetylpyridinium chloride, methylbenzethonium chloride, and 4-picoline dodecyl sulfate.

Serial No. 09/914,279

11. (Previously presented) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

- (a) a metabolizable oil; and
- (b) an emulsifying agent;

wherein said emulsifying agent comprises a cationic detergent and wherein said microemulsion comprises 0.01 to 2.5 % by weight of said emulsifying agent.

12. (Currently amended) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

- (a) a metabolizable oil; and
- (b) an emulsifying agent;

wherein said emulsifying agent comprises an anionic detergent and a non-ionic detergent.

13. (Canceled)

14. (Currently amended) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

- (a) a metabolizable oil; and
- (b) an emulsifying agent;

wherein said emulsifying agent comprises an ionic detergent and a non-ionic detergent, wherein said microemulsion further comprises a biologically active macromolecule adsorbed on the surface thereof, and wherein said macromolecule is an immunological adjuvant selected from the group consisting of a CpG oligonucleotide, alum, a bacterial cell wall component, and muramyl peptide.

15. (Original) The microemulsion of claim 14 wherein said oligonucleotide comprises at least one phosphorothioate bond.

16. (Original) The microemulsion of claim 15 wherein said oligonucleotide comprises at least one peptide nucleic acid bond.

Serial No. 09/914,279

17. (Original) The microemulsion of claim 16 wherein said oligonucleotide comprises a nucleotide sequence selected from the group consisting of SEQ ID NOs: 1-28.

18. (Original) The microemulsion of claim 14 wherein said oligonucleotide comprises a CpG motif flanked by two purines immediately 5' to said motif and two pyrimidines immediately 3' to said motif.

19. (Previously presented) The microemulsion of claim 1 wherein said antigen is from a virus.

20. (Original) The microemulsion of claim 19 wherein the viral antigen comprises a viral subunit.

21. (Original) The microemulsion of claim 19 wherein the virus is selected from the group consisting of hepatitis C virus (HCV), hepatitis B Virus (HBV), herpes simplex virus (HSV), human immunodeficiency virus (HIV), cytomegalovirus (CMV), influenza virus (flu), and rabies virus.

22. (Currently amended) ~~The A-microemulsion of claim 19 comprising microdroplets having an adsorbent surface, said microemulsion comprising:~~

~~(a) a metabolizable oil; and~~

~~(b) an emulsifying agent;~~

~~wherein said emulsifying agent comprises an ionic detergent, wherein said microemulsion further comprises an antigen from a virus adsorbed on the surface thereof, and wherein said antigen is selected from the group consisting of HSV glycoprotein gD, HIV glycoprotein gp120, and HIV p55 gag.~~

23. (Currently amended) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

Serial No. 09/914,279

(a) a metabolizable oil; and

(b) an emulsifying agent;

wherein said emulsifying agent comprises an ionic detergent and a non-ionic detergent,
wherein said microemulsion further comprises an antigen adsorbed on the surface thereof, and
wherein said antigen is from a bacterium.

24. (Original) The microemulsion of claim 23 wherein said bacterium is selected from the group consisting of cholera, diphtheria, tetanus, pertussis, *Helicobacter pylori*, and *Haemophilus influenza*.

25. (Currently amended) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

(a) a metabolizable oil; and

(b) an emulsifying agent;

wherein said emulsifying agent comprises an ionic detergent and a non-ionic detergent,
wherein said microemulsion further comprises an antigen adsorbed on the surface thereof, and
wherein said antigen is from a parasite.

26. (Original) The microemulsion of claim 25 wherein said parasite comprises a malaria parasite.

27. (Withdrawn) A method of inducing an immune response in a host animal comprising administering to said animal the microemulsion of any of claims 1 and 14-26.

28. (Withdrawn) The method of claim 27 wherein said host animal is a mammal.

29. (Withdrawn) The method of claim 28 wherein said mammal is a human.

Serial No. 09/914,279

30. (Withdrawn) A method of immunizing a host animal against a viral, bacterial, or parasitic infection comprising administering to said animal the microemulsion of any of claims 1 and 14-26 in an amount effective to induce a protective response.

31. (Withdrawn) The method of claim 30 wherein said host animal is a mammal.

32. (Withdrawn) The method of claim 31 wherein said mammal is a human.

33. (Withdrawn) A method of inducing a Th1 immune response in a host animal comprising administering to said animal the microemulsion of any of claims 1 and 14-26.

34. (Previously presented) A composition comprising a microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising: (a) a metabolizable oil; and (b) an emulsifying agent; wherein said emulsifying agent comprises an ionic detergent, wherein said microemulsion further comprising a biologically active macromolecule adsorbed on the surface thereof, and wherein the biologically active macromolecule is at least one member selected from the group consisting of a polypeptide, a polynucleotide, a polynucleoside, an antigen, a pharmaceutical, a hormone, an enzyme, a transcription or translation mediator, an intermediate in a metabolic pathway, an immunomodulator, and an immunological adjuvant;

and a microparticle having an adsorbent surface, said microparticle comprising: a polymer selected from the group consisting of a poly(α -hydroxy acid), a polyhydroxy butyric acid, a polycaprolactone, a polyorthoester, a polyanhydride, and a polycyanoacrylate; and a second detergent, which may be the same as or different from said ionic detergent.

35. (Original) The composition of claim 34, wherein said microparticle further comprises a first biologically active macromolecule adsorbed on the surface thereof, wherein the first biologically active macromolecule is at least one member selected from the group consisting of a polypeptide, a polynucleotide, a polynucleoside, an antigen, a pharmaceutical, a hormone, an

Serial No. 09/914,279

enzyme, a transcription or translation mediator, an intermediate in a metabolic pathway, an immunomodulator, and an adjuvant.

36. (Original) The composition of claim 34, wherein said microparticle further comprises a second biologically active macromolecule encapsulated within said microparticle, wherein the second biologically active macromolecule is at least one member selected from the group consisting of a polypeptide, a polynucleotide, a polynucleoside, an antigen, a pharmaceutical, a hormone, an enzyme, a transcription or translation mediator, an intermediate in a metabolic pathway, an immunomodulator, and an adjuvant.

37. (Original) The composition of any of claims 34-36, wherein the microparticle comprises a poly(α -hydroxy acid) selected from the group consisting of poly(L-lactide), poly(D,L-lactide) and poly(D,L-lactide-co-glycolide).

38. (Original) The composition of any of claims 34-36, wherein the microparticle comprises poly(D,L-lactide-co-glycolide).

39. (Original) The composition of any of claims 34-36, wherein the second detergent is a cationic detergent.

40. (Original) The composition of any of claims 34-36, wherein the second detergent is an anionic detergent.

41. (Currently amended) The composition of any of claims 34-36, wherein the second detergent is a ~~nonionic~~ non-ionic detergent.

42. (Original) The composition of any of claims 35-36, wherein the first biologically active macromolecule is an antigen selected from the group consisting of gp120, p24gag, p55gag, and Influenza A hemagglutinin antigen.

Serial No. 09/914,279

43. (Original) The composition of any of claims 35-36, wherein the first biologically active macromolecule is a polynucleotide which encodes gp120.

44. (Previously presented) The composition of claim 36, wherein the second biologically active macromolecule is an immunological adjuvant.

45. (Previously presented) The composition of any of claims 34-36, wherein the immunological adjuvant adsorbed to the microparticle is an aluminum salt.

46. (Original) The composition of any of claims 34-45, further comprising a pharmaceutically acceptable excipient.

47. (Previously presented) The composition of any of claims 34-46, further comprising an unadsorbed immunological adjuvant.

48. (Previously presented) The composition of claim 47, wherein the unadsorbed immunological adjuvant is a member selected from the group consisting of CpG oligonucleotides, LTK63, LTR72, MPL, QS21, Quil A, and an aluminum salt.

49. (Previously presented) The composition of claim 48, wherein the unadsorbed immunological adjuvant is an aluminum salt which is aluminum phosphate.

50. (Withdrawn) A method of delivering a therapeutically effective amount of a macromolecule to a vertebrate subject comprising the step of administering to the vertebrate subject the composition of any of claims 35, 36, 42, 43, 44, 47, or 48.

51-54. (Cancelled)

55. (Currently amended) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

Serial No. 09/914,279

(a) a metabolizable oil; and

(b) an emulsifying agent;

wherein, said emulsifying agent comprises an ionic detergent and a non-ionic detergent and wherein said microemulsion is an oil-in-water emulsion comprising oil microdroplets, substantially all of which are less than 1 micron in diameter.

56. (Currently amended) A microemulsion comprising microdroplets having an adsorbent surface, said microemulsion comprising:

(a) a metabolizable oil; and

(b) an emulsifying agent;

wherein, said emulsifying agent comprises an ionic detergent and a non-ionic detergent and wherein said microemulsion is an oil-in-water emulsion comprising oil microdroplets, at least about 95% of which are less than 1 micron in diameter.

57. (Previously presented) The microemulsion of claim 56 wherein said microemulsion is an oil-in-water emulsion comprising oil microdroplets, at least about 95% of which are less than 0.8 micron in diameter.

58. (Previously presented) The microemulsion of claim 56 wherein said microemulsion is an oil-in-water emulsion comprising oil microdroplets, at least about 95% of which are less than 0.5 micron in diameter.

59. (Previously presented) The microemulsion of claim 55, wherein said metabolizable oil is selected from squalene, squalane, and mixtures thereof.

60. (Previously presented) The microemulsion of claim 59, wherein said ionic detergent is selected from one or more of the following: dioleoyl-3-trimethylammonium-propane, dioleoyl-sn-glycero-3-ethylphosphocholine and dioleoyl-phosphatidic acid.

Serial No. 09/914,279

61. (Currently amended) The microemulsion of claim 55, wherein said emulsifying agent further comprises a ~~nonionic~~ non-ionic detergent is selected from polyoxyethylene sorbitan monoester, polyoxyethylene sorbitan diester, polyoxyethylene sorbitan triester, sorbitan monoester, sorbitan diester, sorbitan triesters, and combinations thereof.

62. (Previously presented) The microemulsion of any of claims 55-61, further comprising an antigen.

63. (Previously presented) The microemulsion of claim 62, wherein said antigen comprises a tumor antigen.

64. (Previously presented) The microemulsion of claim 62, wherein said antigen comprises an antigen derived from a virus.

65. (Previously presented) The microemulsion of claim 62, wherein said antigen comprises an antigen derived from a bacterium.

66. (Previously presented) The microemulsion of claim 62, wherein said antigen comprises an antigen derived from a parasite or a fungus.

67. (Previously presented) The microemulsion of claim 64, wherein said virus is selected from hepatitis C virus (HCV), hepatitis B Virus (HBV), herpes simplex virus (HSV), human immunodeficiency virus (HIV), cytomegalovirus (CMV), influenza virus (flu), and rabies virus.

68. (Previously presented) The microemulsion of claim 65, wherein said bacterium is selected from the group consisting of cholera, diphtheria, tetanus, pertussis, *Helicobacter pylori*, and *Haemophilus influenza*.

Serial No. 09/914,279

69. (Previously presented) A composition comprising the microemulsion of claim 62 and a microparticle having an adsorbent surface, said microparticle comprising: (a) poly(α -hydroxy acid) and (b) a second ionic detergent which can be the same as or different from said ionic detergent.

70. (Previously presented) The microemulsion of claim 62, wherein said ionic detergent is a cationic detergent.